REMARKS:

In this Amendment, claims 1-3, 5-9 and 20 are currently amended; claims 4, 10-19, 21 and 22 are cancelled without prejudice or disclaimer; and claims 23-26 are newly added. The amended and new claims are supported by the original and previously filed claims, and by the specification of the instant application as filed.

More specifically, support for amended claim 6 is found in the instant specification, *inter alia*, at page 39, line 25; support for amended claim 7 is found in the instant specification, *inter alia*, at page 10, lines 12-15; support for amended claim 20 is found in the instant specification, *inter alia*, at page 6, lines 28-29; support for new claims 23 and 24 is found in the instant specification, *inter alia*, at page 6, lines 7-16; support for new claim 25 is found in original claim 9 and in the instant specification, *inter alia*, at page 11, lines 3-5 and 12-15; and support for new claim 26 is found in original claim 2. Thus, no new matter has been introduced into the application by virtue of the amended and new claims. Accordingly, claims 1-3, 5-8, 20 and 23-26 are currently pending in this application.

The Examiner has stated that an abstract of the disclosure is required on a separate sheet for this application. Accordingly, a separate Abstract of the Disclosure is provided herewith in compliance with this requirement.

In addition, as required by the Examiner, the specification has been amended to refer to sequences by a sequence identifier, preceded by "SEQ ID NO:", in the text of the description or claims.

Claim Objections

Applicants respectfully submit that the Examiner's objections to claims 7, 8 and 20 have been overcome by the presently amended claims, which contain the appropriate corrections.

The Written Description Rejections

Claims 1-9, 20 and 21 stand rejected under 35 U.S.C. §112, first paragraph, as allegedly failing to comply with the written description requirement.

Applicants respectfully submit that the presently claimed invention, drawn to an isolated phenotypic modifying genetic sequence (PMGS) of SEQ ID NO:1, or a sequence having at least 80% identity to SEQ ID NO:1, as well as constructs and methods comprising the PMGS of SEQ ID NO:1, is clearly and sufficiently described and exemplified in the instant specification, such that one of skill in the pertinent art would recognize that applicants had possession of the invention as claimed. The specification elucidates that one mode of action for the PMGS to prevent or reduce silencing of a nucleotide sequence proximal to it, is by preventing or reducing methylation (or by promoting demethylation) of the proximal nucleotide sequence. Support for this function of the PMGS is found in the instant specification, *inter alia*, in Examples 1 and 2, and in Figure 3.

The presently amended claims are supported by the description in the specification, which teaches the tomato α-amylase gene promoter PMGS depicted in SEQ ID NO:1 and sets forth the sequence. The specification exemplifies a tomato line, UQ406, in which a transposable genetic sequence Ds has inserted into SEQ ID NO:1, which applicants have identified as a PMGS, (*See, e.g.,* Example 2, page 27; Example 3, page 28), and a *nos:Bar* gene flanked by the PMGS, thus yielding a PMGS that is

proximal to an exogenous nucleic acid in accordance with the invention. Insertion of the Ds transposon into SEQ ID NO:1 prevents Ac transposon-induced silencing of the *nos:Bar* reporter gene. Figure 3 shows that after exposure to AC, the *nos* promoter of the *nos:Bar* reporter gene in Ds is unmethylated when Ds is inserted into SEQ ID NO:1 and the reporter gene is active; however, the reporter gene is methylated and silenced when Ds is inserted into other loci. Further, in the UQ406 line, a demethylation of *nos* promoter sequences (which drive the expression of the *bar* gene) enables expression of the *bar* gene sufficient to confer resistance to selection agent phosphinothricin (PPT), as described in Example 2. In addition, use of the PMGS, such as in the UQ406 tomato line to prevent the silencing of genes in other plant species, is described in the instant specification at page 39, lines 1-5, Example 9. Such disclosure in the instant specification demonstrates that the PMGS of SEQ ID NO:1 is able to prevent silencing of a gene that is inserted proximal to it, and thus silencing is prevented by prevention of methylation or promotion of demethylation of the nucleic acid molecule inserted proximally to the PMGS. Accordingly, functional properties of the PMGS of SEQ ID NO:1 are taught in the instant specification.

In view of the foregoing, Applicants respectfully submit that the presently claimed subject matter is sufficiently described so as to distinguish it from other materials and to demonstrate that Applicants were in possession of the invention as presently claimed at the time of filing. Reconsideration and withdrawal of this rejection under 35 U.S.C. §112, first paragraph, are thus respectfully requested.

The Enablement Rejections

Claims 1-9, 20 and 21 stand rejected under 35 U.S.C. §112, first paragraph, as allegedly failing to comply with the enablement requirement.

The subject matter of the present claims is directed to a PMGS of SEQ ID NO:1; an isolated nucleotide sequence having at least 80% similarity to the PMGS of SEQ ID NO:1 after optimal sequence alignment; a construct containing the PMGS; and methods of increasing or stabilizing expression of a nucleotide sequence, or preventing or reducing silencing of a nucleotide sequence, employing the PMGS of SEQ ID NO:1 in plants. Applicants respectfully submit that the instant specification provides adequate guidance for making and using the claimed invention commensurate with the scope of the presently claimed invention.

Using a transposon system (Example 1, page 27), Applicants have shown that a tomato cell line, UQ406, carries an active *nos:BAR* selectable marker gene due to insertion of a *Ds* transposon into the PMGS of SEQ ID NO:1 (the α-amylase promoter). In the UQ406 line expressing the PMGS of SEQ ID NO:1, the *nos* promoter is only partially methylated, thereby allowing expression of the normally inactive *bar* gene, and the *Not*I site is unmethylated. (Example 2, pages 27-28 and Figure 3). According to the present invention, the α-amylase promoter sequence, which flanks the active *nos:Bar* gene in the UQ406 tomato line, is designated a PMGS. (Example 3, page 28). The PMGS nucleic acid of SEQ ID NO:1, as assessed by the presence of the *Ds* transposable element containing the *nos:Bar* plant selectable marker, is associated with an unmethylated (i.e., active) *Bar* gene, and with the alleviation of transgene silencing in plants, thereby demonstrating function of the PMGS of SEQ ID NO:1. (*See, e.g.*, Example 9). The instant application also teaches that constructs containing the PMGS of SEQ ID NO:1, flanking a plant selectable marker sequence such as *nos:Bar, nos:Luc,* or *nos:GUS*, can be made and used in accordance with the invention to stabilize or initiate expression of a plant nucleic acid sequence in different plant species and varieties. (Examples 9 and 10).

The Examiner has stated that it is unpredictable whether a nucleotide sequence having at least 25% similarity to a promoter sequence would exhibit promoter function, since it is unpredictable whether such a sequence would possess all the particular nucleotides necessary to mediate specific promoter function. In view of the presently amended claims, Applicants respectfully submit that a sequence of at least 80% sequence identity with the claimed PMGS of SEQ ID NO:1 would be reasonably expected by one skilled in the pertinent art to have a function similar to that disclosed for the claimed PMGS.

The Examiner has also stated is unpredictable that a promoter sequence from a plant can alter the expression of a nucleotide sequence in other species. However, at the time of the invention, it would have been a routine matter to select a promoter and combine it with a gene or exogenous nucleotide sequence in an expression construct for expression in plants or plant cells. The mechanics of selecting vectors, promoters and genes and combining them into expression constructs were well known at the time of Applicants' invention.

Those in the art were also aware that isolated promoters from a variety of sources could be used in constructs containing operatively linked heterologous nucleic acid sequences and that a variety of different promoters work to drive the expression of linked sequences in different types of cells. For example, in the Ronemus et al. publication, the authors describe a construct in which a promoter from one kind of plant, i.e., the cauliflower CaMV 35S constitutive promoter, is used to control the expression of an unrelated nucleotide sequence in a different plant, i.e., MET1 cDNA of *Arabidopsis thaliana,* in the Columbia strain of *Arabidopsis*. Also, Gausing et al. (U.S. Patent No. 5,498,832) teach constructs in which functional promoters from several diverse species and sources, e.g., a CaMV promoter, the NOS promoter, a ubiquitin promoter, or a plant

virus promoter) are operably linked to an unrelated nucleotide sequence, namely, all or a portion of the potato α-amylase gene. Such constructs containing a combination of a heterologous promoter and associated nucleotide sequence are used to produce active α-amylase enzymes and transgenic potato plants. Thus, based on the state of the art at the time of Applicants' invention with respect to molecular biological techniques and the design and production of expression constructs, it would have been a matter of routine experimentation to construct a vector containing a promoter from one cell type in combination with a heterologous nucleotide sequence for expression in a cell type different from that of promoter origin.

Given the present claims and guidance provided by the disclosure of the instant specification, no undue experimentation would be required for one having skill in the art to make and use the PMGS of SEQ ID NO:1, a construct harboring this PMGS and an exogenous nucleic acid, and to practice a method of introducing the PMGS into a plant or plant cell so as to modulate (e.g., increase or stabilize) the expression of the nucleic acid sequence in the plant or plant cell.

The law is clear that a single enabled use is sufficient to satisfy the requirements of 35 U.S.C. § 112, first paragraph. *See, Raytheon v. Roper,* 724 F.2d 951 (Fed. Cir. 1983); *In re Gottlieb,* 328 F.2d 1016 (C.C.P.A. 1964); MPEP §2107.02. In view of the exemplification showing the unmethylated state and activation of an exogenous marker gene that is associated with the claimed PMGS of the invention, it is submitted that the invention is enabled as presently claimed. Accordingly, withdrawal of the enablement rejection is respectfully requested.

The Indefiniteness Rejections

Claims 1-9, 20 and 21 stand rejected under 35 U.S.C. §112, second paragraph, as allegedly failing to particularly point out and distinctly claim the subject matter regarded as the invention.

Claims 1, 3, 4, 6 and 20 have been amended or cancelled without prejudice (in the case of claim 9) herein to address the Examiner's concerns regarding the allegedly indefinite recitations in these claims. Regarding the term "modulating" in claims 3 and 6, this term is used and understood by those having routine skill in many areas of biology, including neurobiology, immunology and molecular biology, to indicate changing, influencing or affecting a function or activity, for example, by increasing, enhancing, decreasing, inhibiting, or stabilizing a function or activity, such as gene or gene product expression. The term is also described in the instant specification in connection with expression of the amylase gene as "increasing or stabilising expression of the amylase gene or decreasing or inhibiting the amylase gene." Because this term is art-recognized and routinely used, Applicants respectfully submit that "modulating" as recited in claims 3 and 6 is definite, clear and understood by the skilled practitioner in the pertinent art. Thus, Applicants respectfully request that the indefiniteness rejections be reconsidered and withdrawn.

The Utility Rejection

Claim 20 stands rejected under 35 U.S.C. §101 as allegedly being directed to nonstatutory subject matter. The Examiner states that claim 20, as written, does not sufficiently distinguish over nucleic acids as they naturally exist. In response to this rejection, claim 20 has been amended so as to distinguish over naturally-occurring

products. Accordingly, withdrawal of the rejection under 35 U.S.C. §101 is respectfully requested.

The Three Anticipation Rejections

Claims 1, 3-9 and 20-21 stand rejected under 35 U.S.C. § 102(b) as allegedly being anticipated by WO 96/12813, published May 2, 1996. According to the Examiner, WO 96/12813 "teaches an isolated nucleic acid sequence that increases or stabilizes expression of an α-amylase gene and that has at least 25% similarity to SEQ ID NO:1, and a method of increasing or stabilizing the expression of a nucleotide sequence." The Examiner refers to the nucleic acid sequence set forth in Figure 4 of the cited PCT publication and opines that the sequence of Figure 4 "would inherently modulate the expression of any nucleic acid to which it is operatively linked", as the Figure 4 sequence is a promoter sequence.

It is well established that to anticipate under §102, each and every limitation of a claimed invention must be disclosed in a single reference. *Apple Computer, Inc. v. Articulate Systems, Inc.*, 234 F.3d 14, 57 USPQ2d 1057 (Fed. Cir. 2000); *Brown v. 3M*, 265 F.3d 1349, 60 USPQ2d 1375 (Fed. Cir. 2001).

Applicants respectfully submit that the WO 96/12813 publication does not contain each and every element of the invention as presently amended. The cited publication discloses a promoter for potato α-amylase, identified in SEQ ID NO:1 of WO 96/12813. The sequence of the potato α-amylase promoter and the sequence of the tomato α-amylase PMGS having SEQ ID NO:1 of the present invention are compared in Figure 4 of the instant application and have less than 80% identity at the nucleic acid level. The presently claimed invention is directed to the PMGS of SEQ ID NO:1; isolated nucleotide sequences with 80% or more identity; and constructs and methods comprising

this PMGS. Because the disclosed promoter in WO 96/12813 is clearly distinct from the PMGS of SEQ ID NO:1 as presently claimed, the cited publication neither expressly nor inherently anticipates the presently claimed invention.

In view of the foregoing, withdrawal of the rejection over the WO 96/12813 publication under 35 U.S.C. § 102(b) is respectfully requested.

Claims 1-3, and 6 stand rejected under 35 U.S.C. § 102(b) as allegedly being anticipated by Ronemus et al. (1996, *Science*, 273:654-657), ("Ronemus"). According to the Examiner, Ronemus "teach[es] an isolated nucleic acid sequence that inhibits methylation of a second nucleotide sequence", which would "necessarily increase or stabilize expression of any second proximal nucleotide sequence, including a heterologous α-amylase or *Dem* coding sequence."

Ronemus does not disclose applicants' PMGS of SEQ ID NO:1. Ronemus describes a construct containing a 4.3 kb antisense sequence of MET1 cDNA under the control of a constitutive promoter (CaMV 35S) introduced into a strain of *Arabidopsis* to determine the resulting methylation patterns. Because the cited reference does not contain each and every limitation of the claimed invention, this reference neither expressly nor inherently anticipates applicants' invention as presently claimed. *Apple Computer, Inc. v. Articulate Systems, Inc.*, 234 F.3d 14, 57 USPQ2d 1057 (Fed. Cir. 2000); *Brown v. 3M*, 265 F.3d 1349, 60 USPQ2d 1375 (Fed. Cir. 2001).

In view of the above, withdrawal of the rejection over Ronemus under 35 U.S.C. § 102(b) is respectfully requested.

Claims 1, 4 and 5 stand rejected under 35 U.S.C. § 102(b) as allegedly being anticipated by Gausing et al. (U.S. Patent No. 5,498,832, issued March 12, 1996), ("Gausing"). According to the Examiner, Gausing "teach[es] an isolated nucleic acid encoding an α-amylase (Figures 1 and 2)", which "would inherently increase or stabilize expression of a second proximal nucleotide sequence."

Gausing teaches nucleotide sequences of potato α-amylase genes and their encoded amino acid sequences, as well as vectors which express potato α-amylase enzymes. Gausing does not teach or disclose applicants' PMGS of SEQ ID NO:1, or its use in methods of increasing or stabilizing an exogenous nucleic acid. This cited reference does not contain each and every limitation of the claimed invention. Thus, Gausing neither expressly nor inherently anticipates applicants' invention as presently claimed.

In view of the above, withdrawal of the rejection over Gausing under 35 U.S.C. § 102(b) is respectfully requested.

CONCLUSION:

Should any additional fees be deemed to be properly assessable in this application during its pendancy, or for the timely consideration of this Amendment, the Commissioner is hereby authorized to charge any such additional fee(s), or to credit any overpayment, to Deposit Account No. 08-0219, Order No. 111590-120.

In the event that the Examiner is of the opinion that further discussion is necessary, the Examiner is hereby respectfully requested to telephone the applicant's undersigned attorney at (212) 937-7258.

Respectfully submitted,

HALE AND DORR LLP

Date: December 3, 2003

By: _____
M. Lisa Wilson
Registration No. 34,045

Correspondence Address:
HALE AND DORR LLP
300 Park Avenue
New York, New York 10022
Telephone: (212) 937-7200
Facsimile: (212) 937-7300

ABSTRACT OF THE DISCLOSURE

Nucleic acid molecules capable of modifying phenotypic traits in eukaryotic cells and in particular plant cells are described. The nucleic acid molecules of the present invention are referred to as "phenotype modifying genetic sequences" or "PMGSs" and may be used to increase and/or stabilise or otherwise facilitate expression of nucleotide sequences being expressed into a translation product. Alternatively, PMGSs may be used to down regulate by, for example, promoting transcript degradation via mechanisms such as co-suppression. In plant and non-plant cells, the PMGSs of the invention may also be used to inhibit, reduce or otherwise down regulate expression of a nucleotide sequence, such as a eukaryotic gene, including a pathogen gene, the expression of which results in an undesired phenotype.